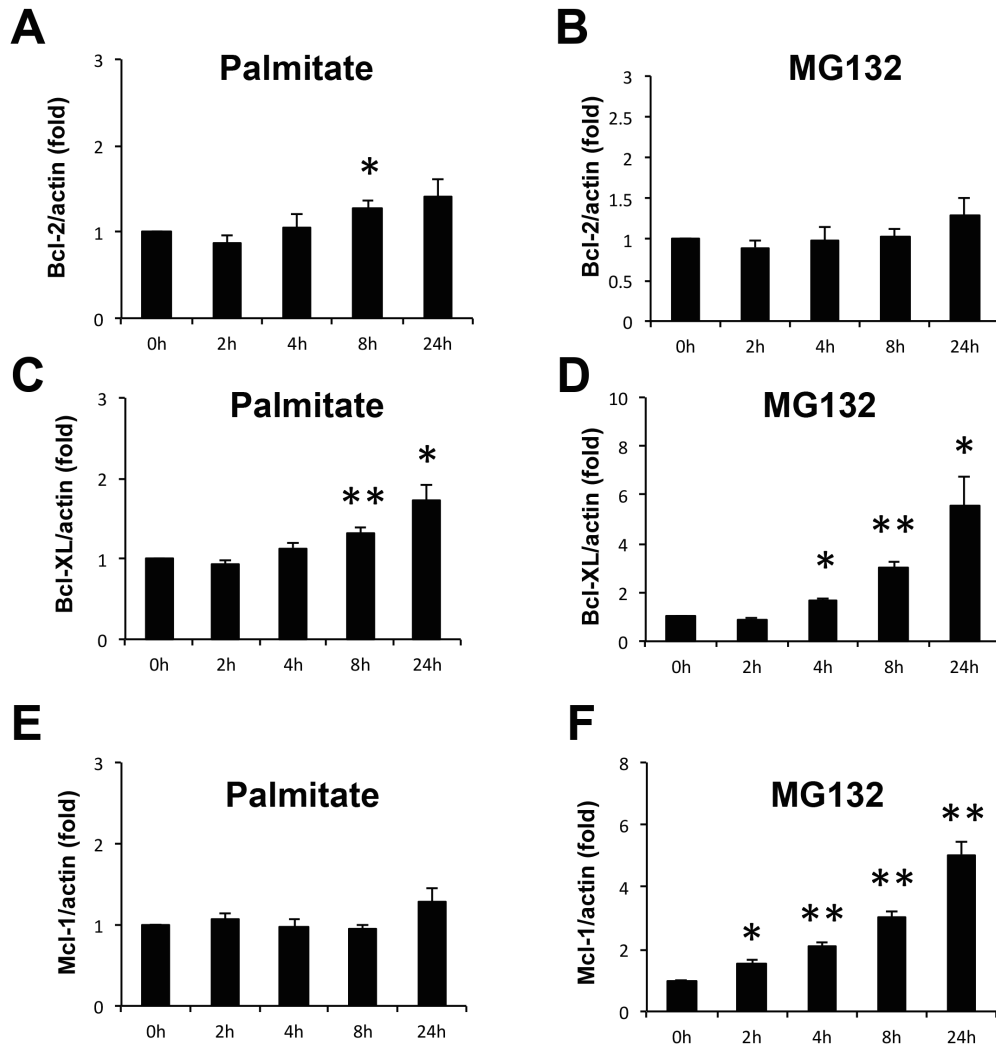


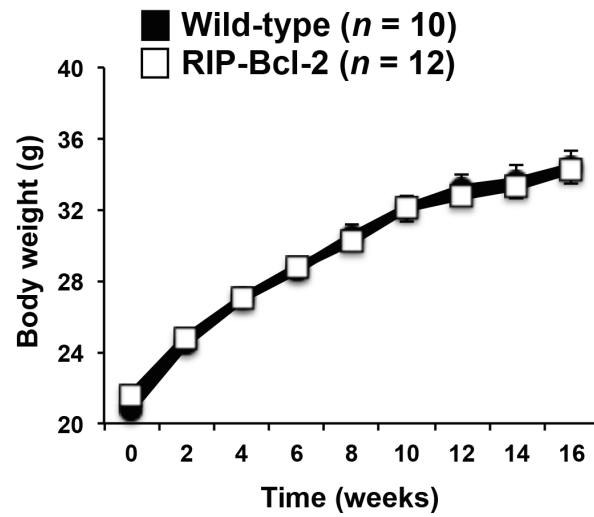
Supplemental Data

Lipotoxic stress induces pancreatic β -cell apoptosis through modulation of Bcl-2 proteins by the ubiquitin-proteasome system.

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Supplementary Figure 1. Palmitate and MG132 do not reduce mRNA levels of pro-survival proteins in β -cells. (A,C,E) MIN6 cells were treated with 0.5 mM palmitate (A, C, E) or 10 μ M MG132 (B, D, F) and expression of pro-survival Bcl-2 (A,B), Bcl-XL (C,D) and Mcl-1 (E,F) was measured by qPCR. Results are the means \pm SEM of 3 independent experiments, and represented as fold induction compared to the untreated control samples. *P < 0.05, **P < 0.01.



Supplementary Figure 2. Six week-old male wild-type and RIP-Bcl-2 mice were fed a high fat diet for 16 weeks and body weights measured.

Sample ID	Age	Gender	BMI	T2D
SVI-032-10	40	F	24.2	-
SVI-034-10	31	M	21.2	-
SVI-021-10	70	F	22.6	-
SVI-021-11	53	F	34.5	-
SVI-007-11	47	M	38.9	+
SVI-035-10	53	M	30.1	+
SVI-023-14	61	F	32	-
SVI-024-11	47	M	38.9	-

Supplementary Table 1. List of human samples used in the study.

Probe	Catalogue number (Applied Biosystems, Foster City, CA, USA)
ATF4 (mouse)	Mm00515325_g1
Chop (mouse)	Mm01135937_g1
Bip (mouse)	Mm00517690_g1
Bcl-2 (mouse)	Mm00477631_m1
Bcl-XL (mouse)	Mm00437783_m1
Mcl-1 (mouse)	Mm00725832_s1
PUMA (mouse)	Mm00519268_m1
β-actin (mouse)	Mm00607939_s1

Supplementary Table 2. List of probes used for qPCR. Real-time PCR was performed using the Rotor-Gene RG-3000 machine (Corbett Research; Qiagen, Hilden, Germany) and the TaqMan PCR Master Mix (AmpliTaq Gold with GeneAmp kit; Applied Biosystems) in 20 µl reaction volumes.

Antibody	Company	Reference	Dilution
Ubiquitin	Cell Signaling, Danvers, MA	#3933	1/1000
Cleaved Caspase-3	Cell Signaling, Danvers, MA	#9661	1/500
Chop	Santa Cruz Biotechnology, CA	sc-575	1/500
Bcl-2	Cell Signaling, Danvers, MA	#2870	1/1000
Bcl-XL	Cell Signaling, Danvers, MA	#2764	1/500
Mcl-1	Cell Signaling, Danvers, MA	#5435	1/1000
β-Actin	Santa Cruz Biotechnology, CA	sc-7210	1/5000
HRP-conjugated anti-rabbit IgG	Millipore, Temecula, CA	12-348	1/5000
HRP-conjugated anti-mouse IgG	DAKO, Denmark	P0260	1/10000

Supplementary Table 3. List of antibodies used for Western blot analysis.